

# Seed Production & Quality Management



## *Genetic Variation & Plant Breeding*

### Plant Breeding

The art and science of improving the heredity of economically important plant traits for the benefit of man

- Art : breeder's "eye"
- Science : applied genetics

Plant breeding has a long time horizon (10-20 years)

Plant breeding must constantly re-evaluate its goals ('moving target')

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## *Genetic Variation & Plant Breeding*

### Plant Breeding

Requires knowledge of:

- Anatomy, morphology, physiology, biochemistry, taxonomy, genetics, economics, biometry
- Interactions between plants and their total environment
- Includes climate, insects, disease, soils, plant community, production technologies, etc.

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## *Genetic Variation & Plant Breeding*

### Key issues in successful Plant Breeding

- Presence of and/or possibility for development of genetic variation
- Ability to select desirable genotypes

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## *Genetic Variation & Plant Breeding*

### How is genetic variation developed

- Domestication
- Collection & Introduction of germplasm
- Crosses (intervarietal, distant, somatic)
- Mutations
- Polyploidy
- Somaclonal
- Genetic engineering

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## *Genetic Variation & Plant Breeding*

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## *Genetic Variation & Plant Breeding*

### Main stages in Plant Breeding History

- Domestication (~10,000 until 1,000 BC)
- Empirical Breeding
  - grower's selection (until today)
  - professional occupation (until 19<sup>th</sup> century)
- Modern / Science-based Breeding (after 19<sup>th</sup> century)
  - classical breeding
  - modern approaches

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## *Genetic Variation & Plant Breeding*

### Plant Domestication

Started in a small number of regions (Centres of Origin – Vavilov)



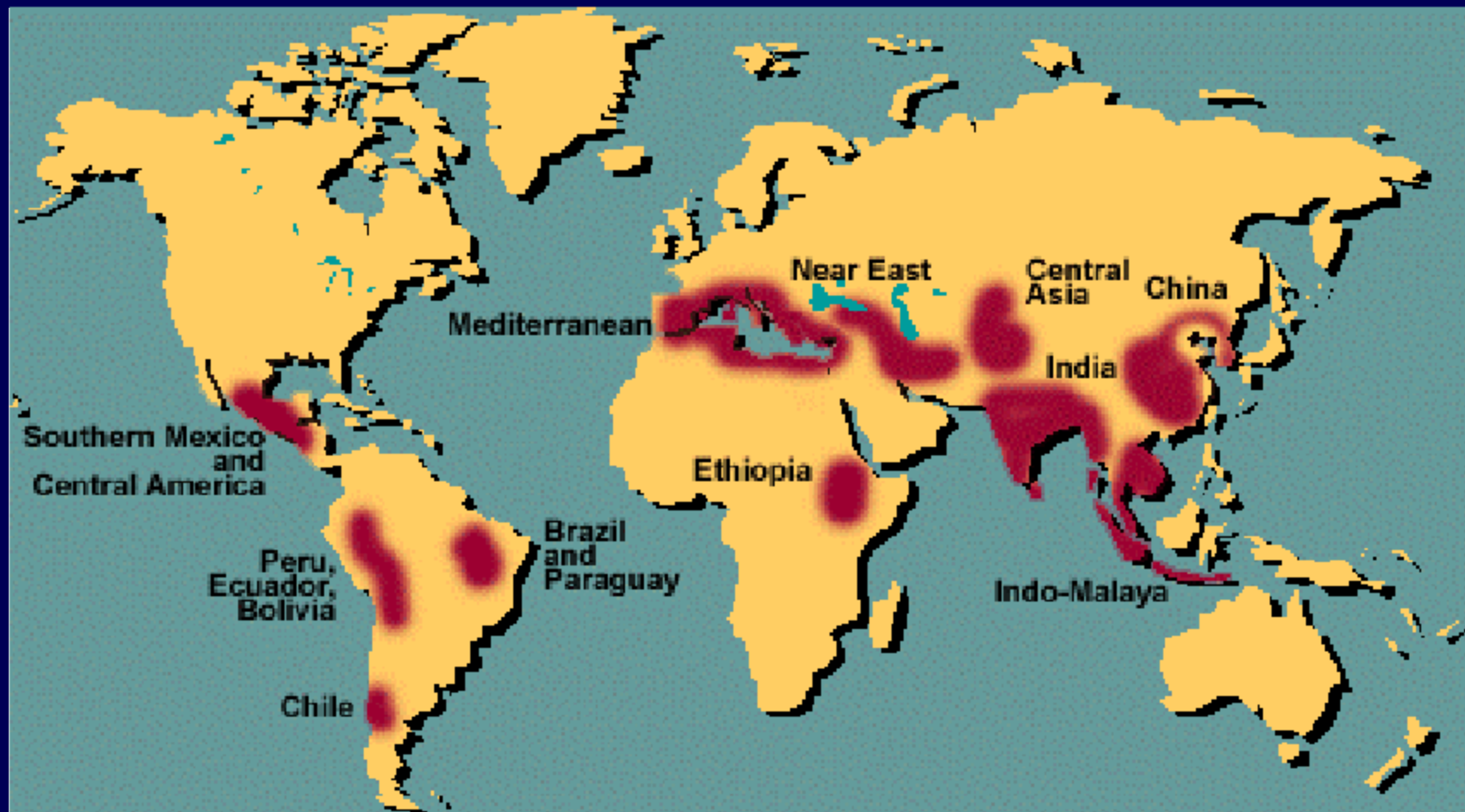
- Initially referred to a few species such as wheat, barley, maize, potato, beans, etc
- Traits that played important role in the process were nutritive value, harvestability and storability

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*Genetic Variation & Plant Breeding*

Centres of origin/diversity (Vavilov)





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## *Genetic Variation & Plant Breeding*

### Process of Plant Domestication

- Early domestication used form of Mass Selection
- Took advantage of 'natural' mutations
- Seed saved enriched the following year's crop with desirable traits
- After 'several' cycles, crops fixed or purged relevant traits/genes

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## *Genetic Variation & Plant Breeding*

### Effects of Plant Domestication on the Phenotype

- Domestication at first affected traits related to:
  - Harvestability, storability, chemistry, adaptability
- Purged undesirable traits early ( ie.shattering rachis)
- Fixed desirable traits early ( ie. thin seed coats)
- Relatively few, but important forms were developed
- Created a 'uniform' series of populations for the traits under selection

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## *Genetic Variation & Plant Breeding*

### Effects of Plant Domestication on the Phenotype

Teosinte (top)

Modern Hybrid Corn (bottom)



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## *Genetic Variation & Plant Breeding*

### Effects of Plant Domestication on the Phenotype (cont)



Undomesticated germplasm

Modern varieties

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## *Genetic Variation & Plant Breeding*

### Effects of Plant Domestication on the Genotype

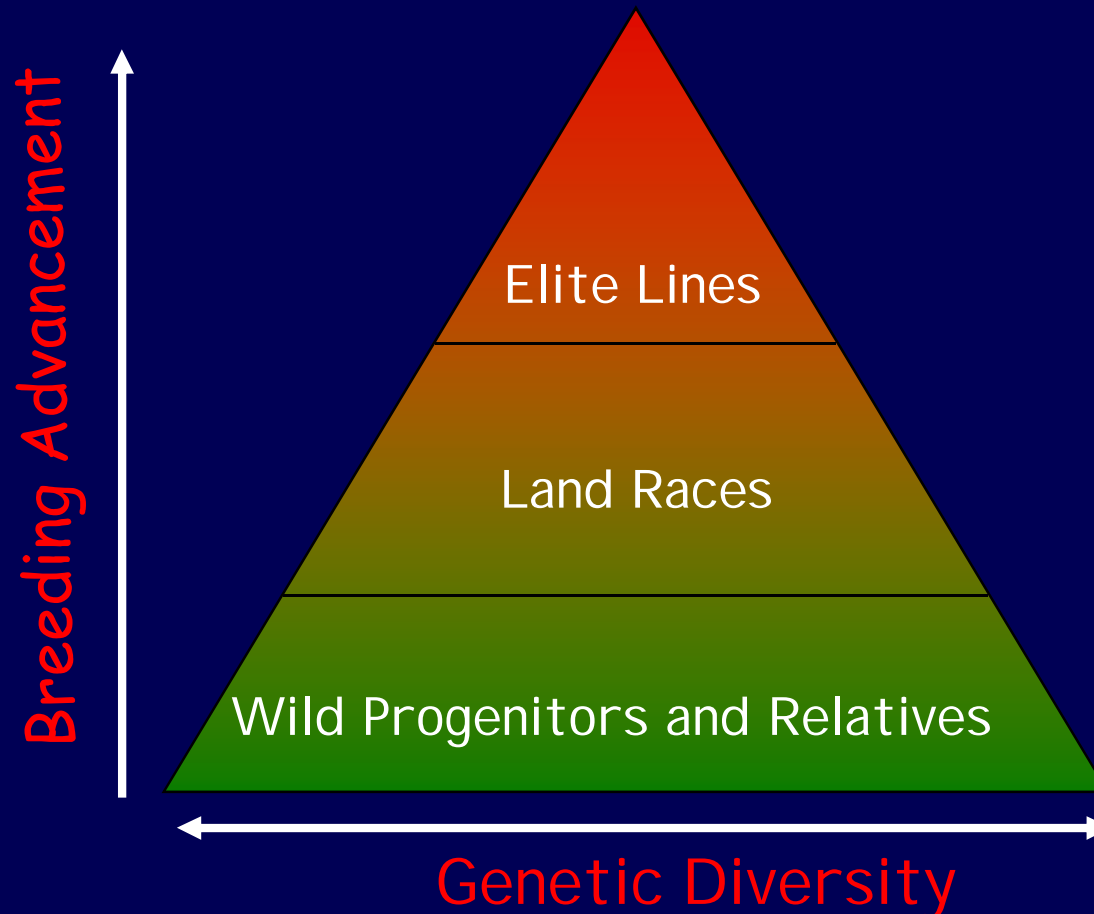
- Narrowed genotypic variability for traits under selection but maintained variability for non-selected traits
- Inadvertently purged genes controlling traits not under intentional selection
- Fixed genes controlling traits under strong selection (non-shattering rachis)
- Relatively few genes selected but indirectly affected more due to linkage
- Created a heterogeneous series of populations adapted to local environments (land races)

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## *Genetic Variation & Plant Breeding*

Domestication and Modern Plant Breeding has Narrowed the Gene Pool and Caused Biodiversity Loss



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## *Genetic Variation & Plant Breeding*

### Domestication Summary

- Gains: allowed development of civilization, greater yields, increased usable biomass, fixed desirable traits, purged undesirable traits, increased uniformity
- Losses: reduced adaptability to variable environments, increased susceptibility to stress, reduced competitive ability, reduced genetic variation, increased uniformity

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## *Genetic Variation & Plant Breeding*

### Empirical Plant Breeding

- Grower's empirical selection

Gave rise to the landraces ie. regionally adapted populations endowed with large genetic variation

- Professional occupation

Systematic breeding as for example in flowering and ornamentals, development of the pedigree method in sugarbeet, production of the first artificial hybrid



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*Genetic Variation & Plant Breeding*

## Science-based Plant Breeding



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## *Genetic Variation & Plant Breeding*

### Modern Plant Breeding

- A particularly significant contribution (50%) to the overall increase in crop production
- Production of highly homogeneous varieties
- Based on classical approaches (Mendelian – Quantitative genetics ) and Biometry
- Modern biotechnological methods (*in vitro* culture – Molecular breeding - Genetic engineering) have already allowed for a dramatic increase in plant breeding program's efficiency

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## *Genetic Variation & Plant Breeding*

### Gains due to Modern Plant Breeding

- Total productivity increased
- Production area expanded
- Hybrid cultivars developed
- Resistance to biotic and abiotic stress causal agents manipulated
- Food quality improved
- Crops designed for adaptability to mechanical harvest

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## *Genetic Variation & Plant Breeding*

### Losses due to Modern Plant Breeding

- Loss of crop genetic diversity
- Loss of ecological diversity
- Displacement of locally adapted cultivars
- Environmental contamination
- Alteration of biomass distribution
- Reduced competitive ability
- Loss of beneficial gene combinations

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## *Genetic Variation & Plant Breeding*

### Summary of Modern Plant Breeding Efforts

- Shifted genetic structure of crops from one that favored broad (horizontal) selection of traits across different levels of system function to one that selects heavily toward a narrow (vertical) set of traits without regard to system level functions
- Typically, modern cultivars are produced as large homogeneous populations that are artificially supplied system level support

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## *Genetic Variation & Plant Breeding*

### Sustainability

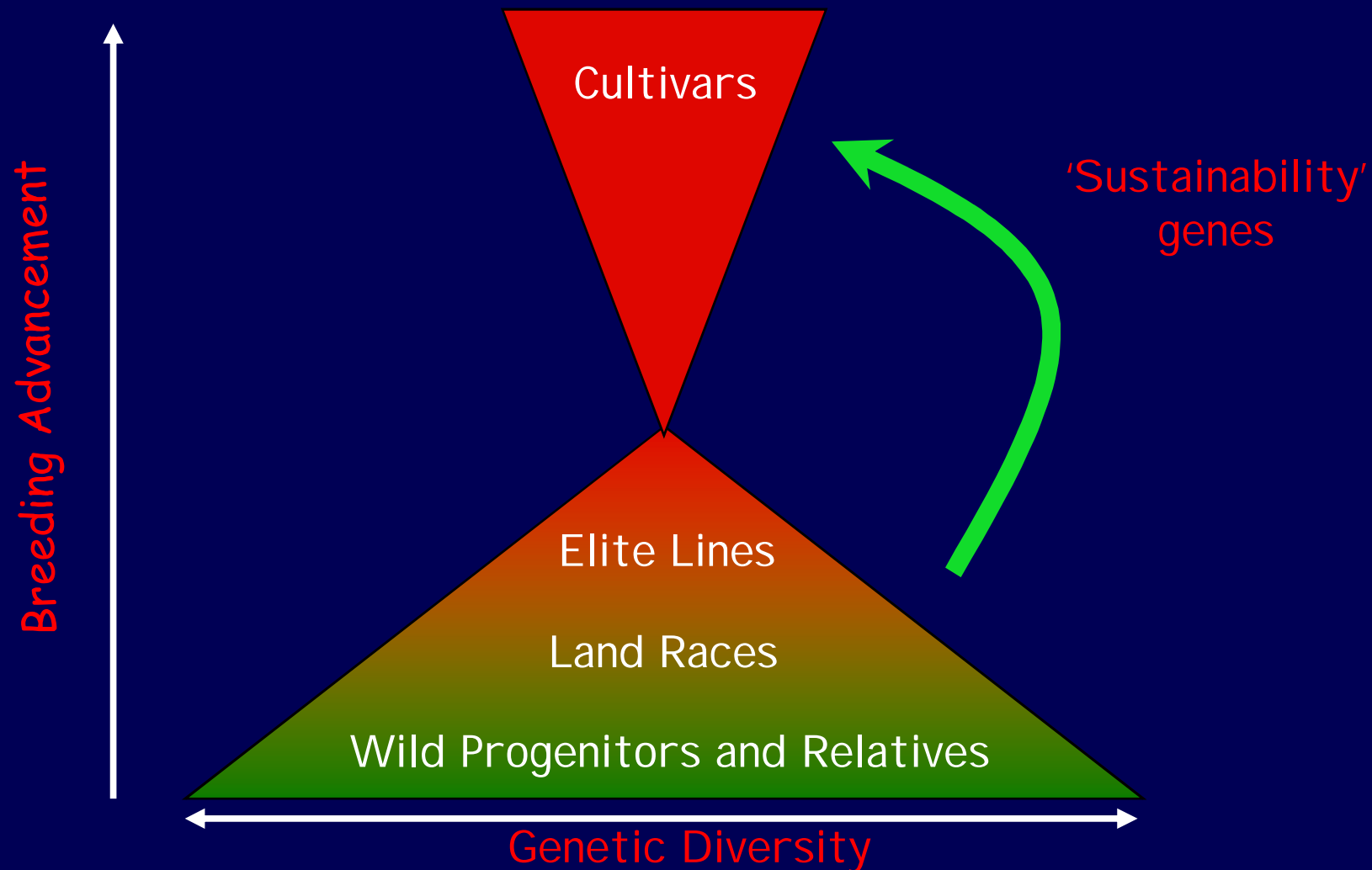
- Ability to meet one generation's needs without compromising future generation's ability of doing the same
- Includes environmental, societal, and economic issues
- Plant breeders are critical players in the development of sustainable production systems paired with high yielding cultivars

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## *Genetic Variation & Plant Breeding*

### Genetic Structure of New Introductions



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## *Genetic Variation & Plant Breeding*

### Traits Related to Sustainability

- Water use efficiency and resistance to drought
- Nutrient uptake efficiency
- Host plant resistance to pests and diseases
- Competitive ability
- Ability to benefit from synergisms across system levels
- Soil conditioning
- Plasticity across stress and environments



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*Genetic Variation & Plant Breeding*

How is variation developed ?

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## *Genetic Variation & Plant Breeding*

### Sources of germplasm (variation)

- Wild species and primitive forms of crops nurtured in primary centers of diversity where the species are in competition with other natural elements of the environment
- Plant migrants nurtured in secondary centers of culture where their diversity might have been augmented due to new biological and physical stresses acting as selective factors
- Products of plant breeding, including induced polyploidy, mutations, and the combining of multiple traits into useful breeding lines

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## *Genetic Variation & Plant Breeding*

### Conservation of Genetic Diversity

*In situ* → In the initial environment

- protected areas
- traditional cultures
- botanical gardens, orchards

*Ex situ* → Outside the initial environment

- botanical gardens
- Gene banks

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## *Genetic Variation & Plant Breeding*

### Conservation *in situ*

#### Advantages

- Less costly
- Continued evolution and adaptation to the environmental changes

#### Disadvantages

- Inadequate control
- Absence of evaluation

### Conservation *ex situ*

#### Advantages

- Well organized
- Accessible for research
- Good documentation and evaluation

#### Disadvantages

- Costly installations
- Absence of evolution
- Genetic drift in small populations

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*Genetic Variation & Plant Breeding*

## Gene (or germplasm) Banks

*The shelter of variation to be used in Modern Plant Breeding*

- Obtaining the germplasm
- Maintenance of germplasm
- Assessment of germplasm
- Documentation of germplasm
- distribution of germplasm

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## *Genetic Variation & Plant Breeding*

### The concept of Gene Pool

*(Harlan and de Wet, 1971)*

Gene pool consists of all the genes present in all such individuals which hybridize or can hybridize with each other. In this sense it describes a concept similar to germplasm

Germplasm resources are classified in various "Gene pools" depending on the ease of hybridization ie. on how readily genes are transferred between its members

Three main categories can be identified:

- Primary gene pool
- Secondary gene pool
- Tertiary gene pool

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## *Genetic Variation & Plant Breeding*

### Gene Pools

Primary gene pool (GP<sub>1</sub>): it refers to the level of botanical species and includes landraces, local cultivars, ecotypes as well as weed and possibly wild subspecies

- Full crossability, absence of sterility problems, normal gene segregation
- Most extensively used by programs that aims at broadening the genetic base
- Most breeders are mainly or entirely using this category

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## *Genetic Variation & Plant Breeding*

### Gene Pools (cont)

Secondary gene pool (GP<sub>2</sub>): members of this gene pool hybridize with those of the primary gene pool with some to considerable difficulty, the hybrids are only partially, if at all, fertile and less of reduced vigor

It includes related species (but not all) in a genus as well as some species outside the genus

Gene transfer from the second to the primary gene pool are possible but usually difficult. Members of this category are often used in breeding programs



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## *Genetic Variation & Plant Breeding*

### Gene Pools (cont)

Tertiary gene pool ( $GP_3$ ): it includes distantly related species in different genera or very little related species of the same genus

Species belonging to this group cross with members of  $GP_1$  with considerable to great difficulty and hybrids, if produced, are invariably sterile. Gene transfers to  $GP_1$ , thus are very difficult and require special techniques (embryo rescue, ovule culture, bridging, chromosome doubling)

Gene transfers from  $GP_3$  to  $GP_2$  are relatively easier.  $GP_3$  is only occasionally used in breeding programs

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## *Genetic Variation & Plant Breeding*

### Exploitation of germplasm in Plant Breeding

- Direct supply (new crops-varieties)
- Incorporation of monogenic traits (backcrossing)
- Quantitative traits
  - products of direct hybridization (potato, alfalfa)
  - incorporation with classical breeding methodology
  - incorporation through molecular approaches
  - incorporation through broad genetic base pre-breeding populations

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## *Genetic Variation & Plant Breeding*

### The main steps in Plant Breeding

- Induction of variation by creation of crosses or by variation-inducing treatment
- Selection of desired traits in new varieties
- Propagation and multiplication of breeding lines

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*Genetic Variation & Plant Breeding*

## Breeding for Organic Agriculture

### Basic principles in Organic Agriculture

- Closed production cycles
- Natural self regulation
- Biodiversity

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## *Genetic Variation & Plant Breeding*

### Organic Breeding (cont)

#### Basic principles in Organic Plant Breeding

- Natural reproductive ability of plants
- Ability to adapt to organic conditions
- Genetic diversity with respect for natural species authenticity and species characteristics

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## *Genetic Variation & Plant Breeding*

### Development of varieties suited to organic agriculture

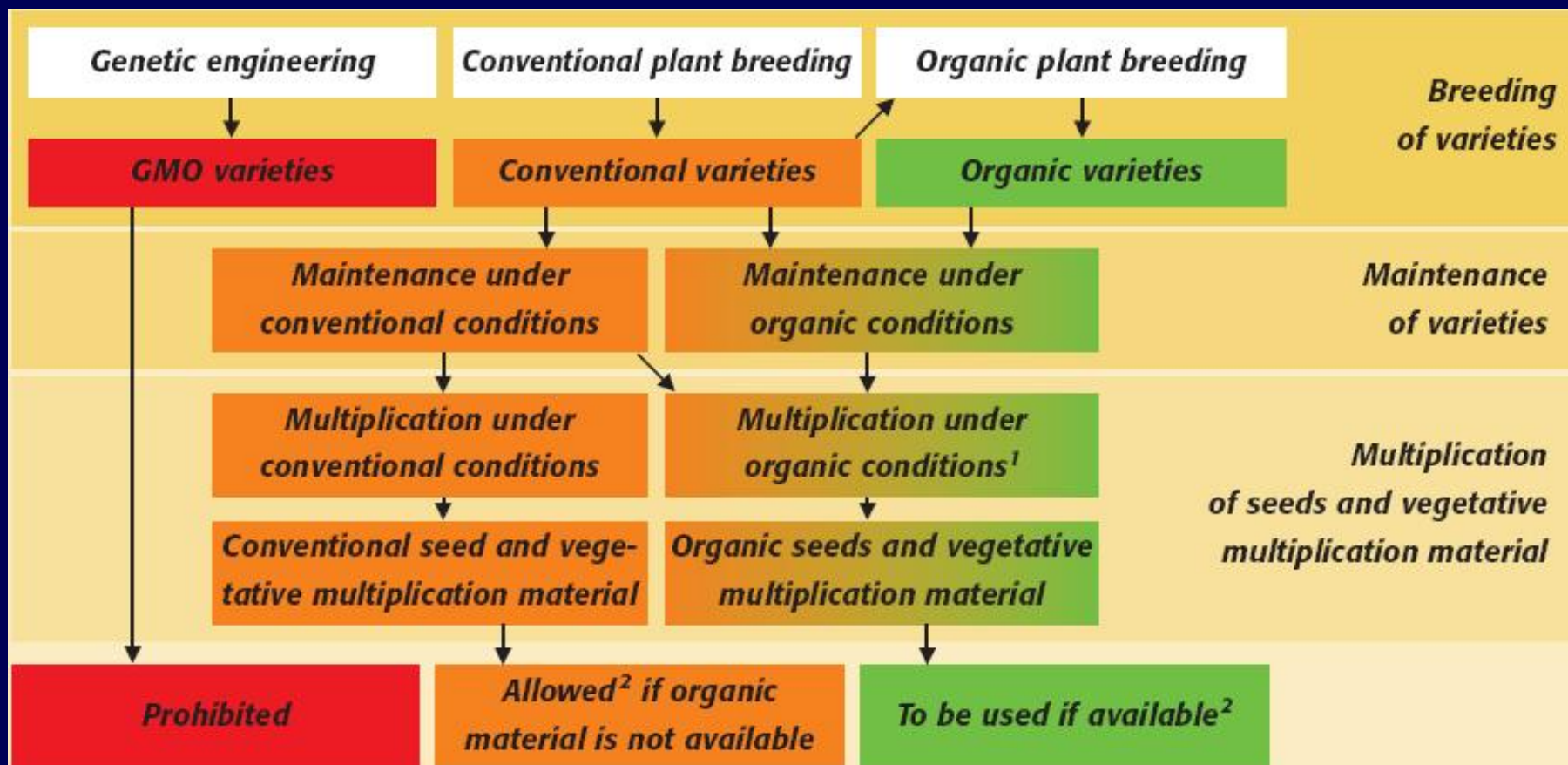
- Optimal adaptation to local environment and nutrient dynamics
- Nutrient efficiency
- Durable resistance and tolerance against pests and diseases
- Yield stability
- Storability
- Nutritional and sensorical quality

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## *Genetic Variation & Plant Breeding*

### Plant Breeding Maintenance and Multiplication



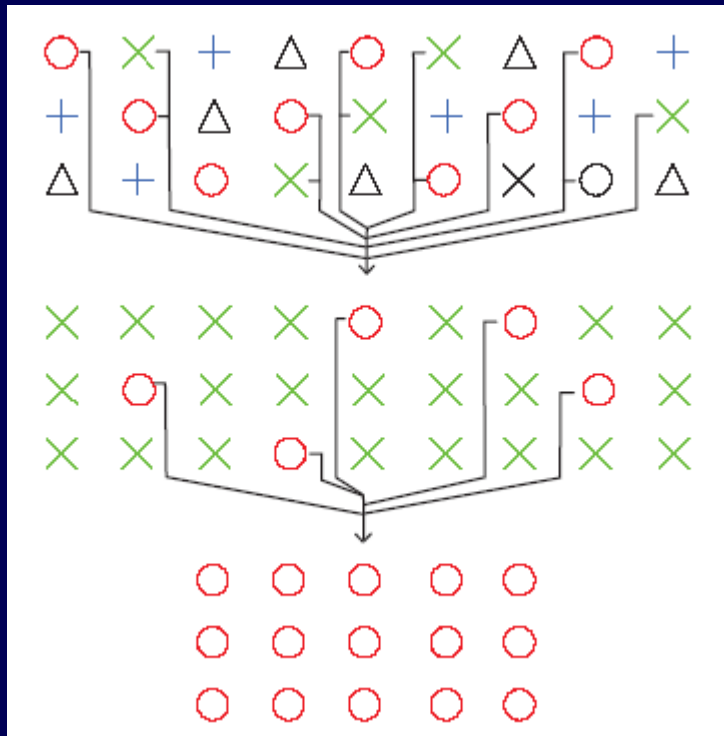
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## *Genetic Variation & Plant Breeding*

### Techniques used in Organic plant breeding

#### Mass selection



Easy and quick method to develop superior cultivars from locally adapted germplasm

Cultivar is developed as a genetic mixture of pure lines; results in higher adaptability and stability of yield

Possess phenotypic uniformity despite having considerable hidden genetic variation

Not effective with low  $h^2$  traits

Population cannot realize maximum potential displayed by the 'best' pure line, due to bulking



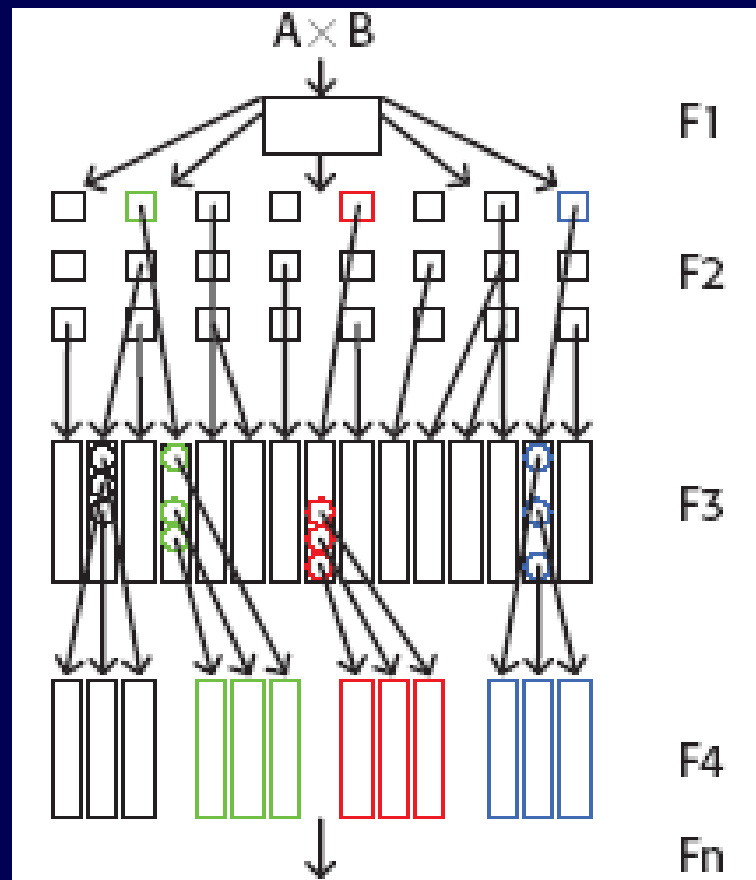
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## *Genetic Variation & Plant Breeding*

### Techniques used in Organic plant breeding (cont)

#### Pedigree selection



Combining the desirable traits of two or more genotypes in a new improved variety or inbred line

Typically, one parent is already an improved locally adapted line while the other brings one or more desirable traits that are often well-expressed

Eliminates unpromising material at early stages

Multiple families (from different F2 individuals) are maintained yielding different gene combinations with common phenotype

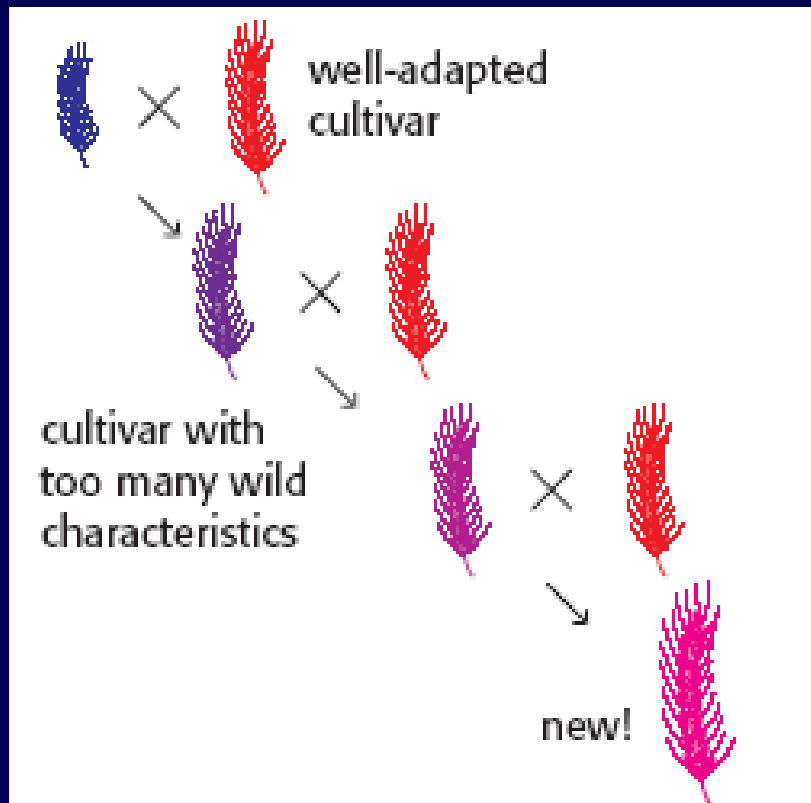
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## *Genetic Variation & Plant Breeding*

### Techniques used in Organic plant breeding (cont)

#### Backcrossing



Designed to recreate the genotype of the recurrent parent plus the desired allele(s) from the other parent (donor parent; non-recurrent parent)

Trait of interest should be simply inherited, if possible and easily phenotyped

BC-derived lines are expected to be well-adapted to the environment in which they will be grown - progeny need less extensive field testing

Provides breeder with high degree of genetic control

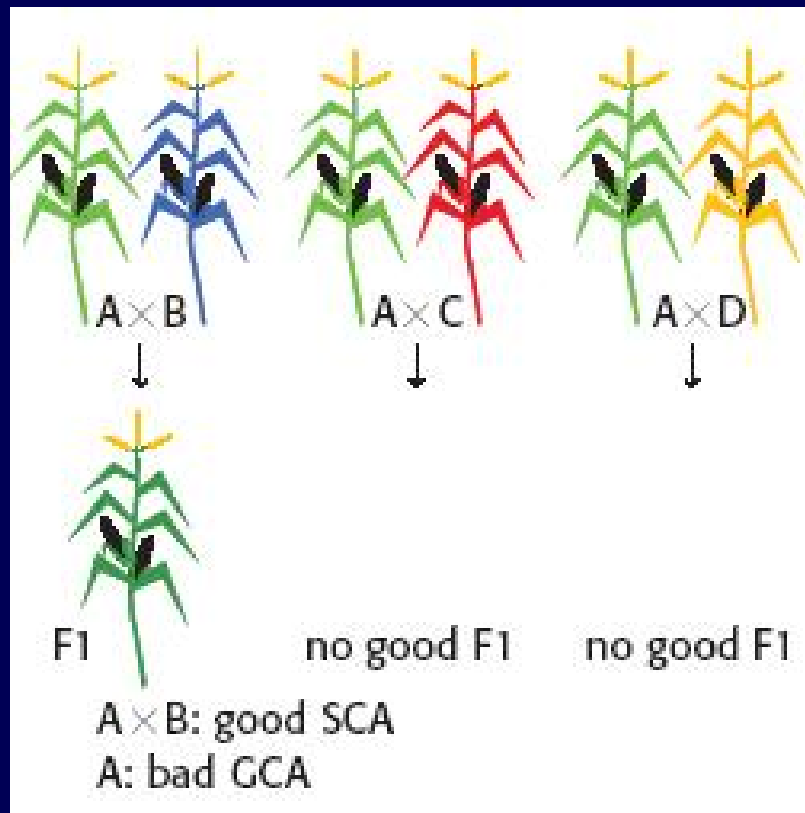
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## *Genetic Variation & Plant Breeding*

### Techniques used in Organic plant breeding (cont)

#### Test crosses



Promising parental genotypes are crossed with a number of other known genotypes and progeny are assessed for desired characteristics to determine GCA and SCA

Mostly used for asexually propagated plants and hybrids

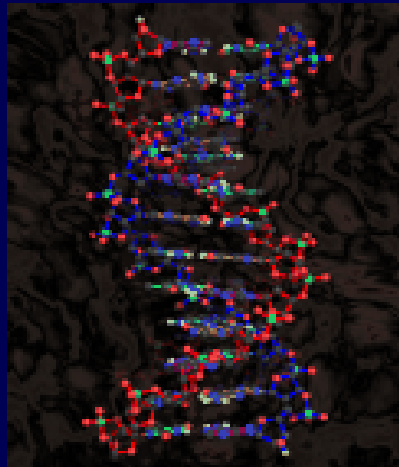
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## *Genetic Variation & Plant Breeding*

### Techniques for selection at the DNA level

#### Marker-assisted selection (MAS)

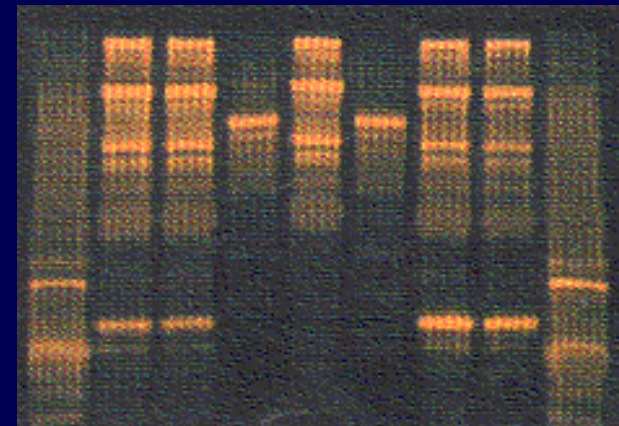


DNA isolation



DNA  
treatment

↑  
*RFLPs*  
*RAPDs*  
*AFLPs*  
*SSRs*  
*SNPs*



Gel electrophoresis  
for the separation  
of DNA fragments

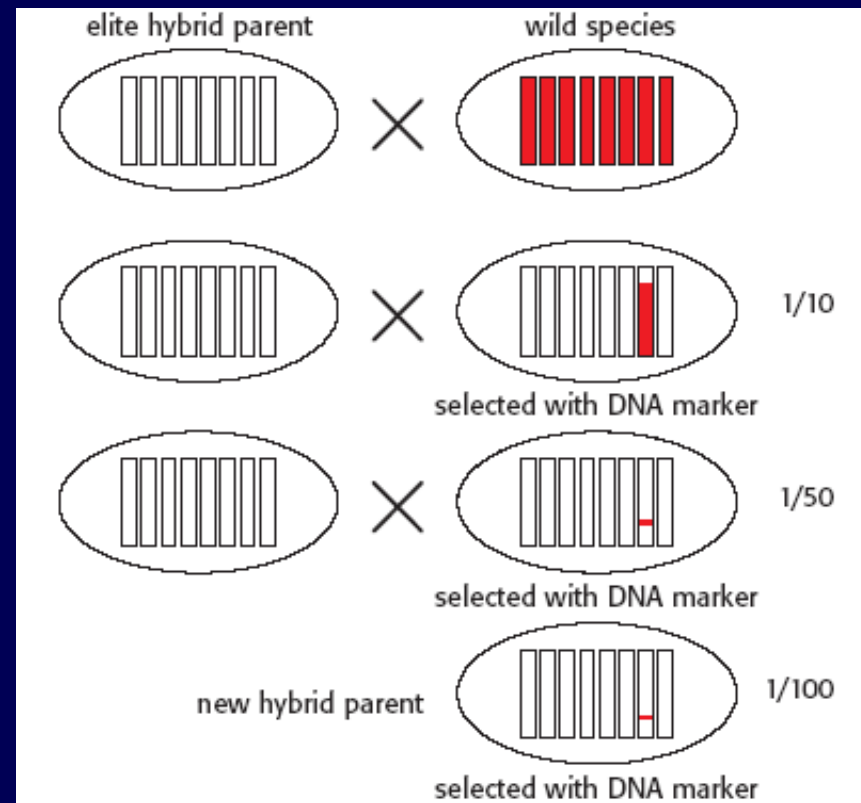
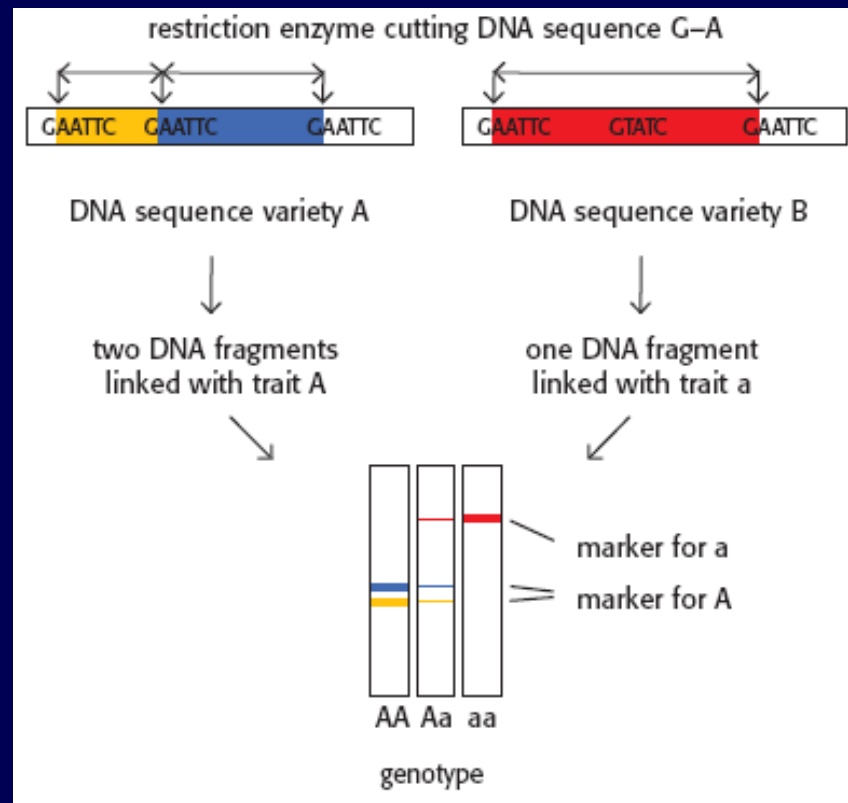
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## *Genetic Variation & Plant Breeding*

### Techniques for selection at the DNA level

#### Marker-assisted selection (MAS)



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## Genetic Variation & Plant Breeding

### Judgment of Techniques for Organic Plant Breeding

<i>Inducing variation</i>	<i>Smallest living entity</i>		
	<i>Plant</i>	<i>Cell</i>	<i>DNA</i>
<i>Combination breeding, Crossing varieties, Bridging cross, Repeated back-crossing, Cutted/grafted style, Temperature treatment of the style</i>	↑	↑	↑
<i>F1 hybrid breeding, Unradiated mentor pollen technique</i>	↗	↑	↑
<i>Ovary/embryo culture, «In vitro» pollination, Anther/Microspore culture</i>	↘	↑	↑
<i>Polyploidisation, Somaclonal variation</i>	↘	↗	↑
<i>Hybridisation for CMS without restorer gene, Radiated mentor pollen technique</i>	↓	↗	↑
<i>Protoplast fusion</i>	↓	↘	↑
<i>Genetic engineering (is already prohibited)</i>	↓	↓	↓

Colours and arrows give the «degree of suitability»: ↑ = no problem, ↗ = ok, ↘ = not suitable but provisionally allowed, ↓ = not suitable.

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## Genetic Variation & Plant Breeding

### Judgment of Techniques for Organic Plant Breeding (cont)

<b>Multiplication</b>	<i>Plant</i>	<i>Cell</i>	<i>DNA</i>
<i>Generative multiplication</i>	↑	↑	↑
<i>Vegetative multiplication</i>	↑	↑	↑
<i>Apomixis</i>	↘	↑	↑
<i>Meristem culture</i>	↘	↗	↑
<i>«In vitro» multiplication, Somatic embryogenesis</i>	↓	↗	↑
<b>Selection</b>	<i>Plant</i>	<i>Cell</i>	<i>DNA</i>
<i>Mass selection, Pedigree selection, Site determined selection, Change in environment, Change in sowing time, Ear bed method</i>			
<i>Test crosses</i>	↑	↑	↑
<i>«In vitro» selection</i>	↓	↗	↑
<i>Marker-assisted selection</i>	↓	↗	↑

Colours and arrows give the «degree of suitability»: ↑ = no problem, ↗ = ok, ↘ = not suitable but provisionally allowed, ↓ = not suitable.

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*Genetic Variation & Plant Breeding*

Discussion on the papers



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## *Genetic Variation & Plant Breeding*

### Breeding for abiotic stress

- Potential yield/ stress environment
- Adapted/stable genotype
- Target population of environments/ Selection environment
- Multi-environment trials/ Managed stress trials
- Forms of GxE
- Indirect selection response theory

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## *Genetic Variation & Plant Breeding*

### Stress, genetic potential and adaptation

- A genotype that cannot realize its potential due to biotic and/or abiotic constraints is called stressed. A stress environment exerts stress in comparison to an optimal environment.
- Yield potential (no stress of any type/ ceiling); yield of genotype growing in an environment to which it is *adapted*, soil-nutrients and water are non-limiting, other (biological) stresses are controlled
- Adaptation: growth and development of genotype that is well matched to the environment/ genotype has high *fitness* under these conditions

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## *Genetic Variation & Plant Breeding*

### Genotype by environment interaction (GxE or GE)

- Genotypes respond differently across a range of environments i.e., the relative performance of varieties depends on the environment

$$P = G + E + GE$$

$$V_P = V_G + V_E + V_{GE}$$

- Genotype by environment interactions are common for most quantitative traits of economic importance
- Advanced breeding materials must be evaluated in multiple locations for more than one year

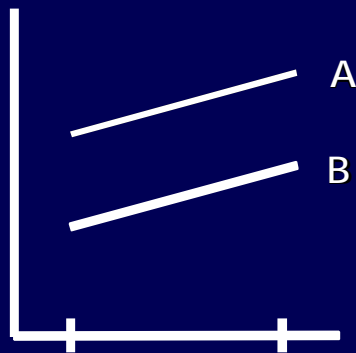
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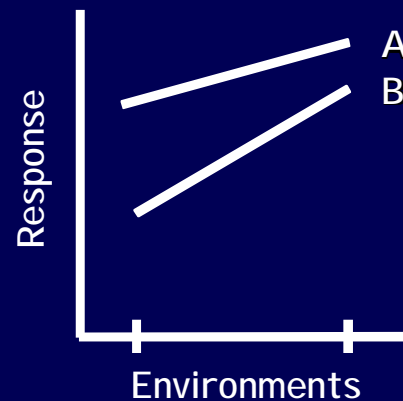
## *Genetic Variation & Plant Breeding*

### Types of GEI

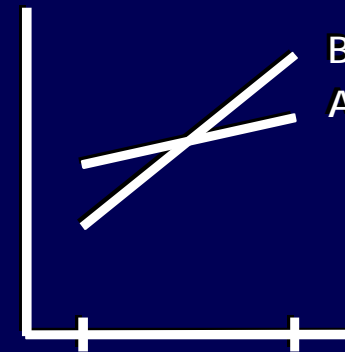
No interaction



No rank changes, but interaction



Rank changes and interaction



- Interaction may be due to:
  - heterogeneity of genotypic variance across environments
  - imperfect correlation of genotypic performance across environments

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## *Genetic Variation & Plant Breeding*

### The GEI challenge

- Environmental effect is the greatest, but is irrelevant to selection
- Many statistical approaches consider all of the phenotypic variation (i.e. means across environments), which may be misleading
- Analysis is essential for the characterization of GEI
- "GE Interaction is not merely a problem, it is also an opportunity" (Simmonds, 1991). Specific adaptations can make the difference between a good variety and a superb variety

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## *Genetic Variation & Plant Breeding*

### The GEI challenge

- Some environmental variation is predictable
  - can be attributed to specific, characteristic features of the environment
  - e.g. soil type, soil fertility, plant density
- Some variation is unpredictable
  - e.g. rainfall, temperature, humidity

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## *Genetic Variation & Plant Breeding*

### Stress environment

- Moisture (drought)
- Temperature (heat, chilling, freezing)
- Soil-Chemical (nutrients, minerals)
- Radiation
- All these factors will cause stress, observable at the phenotypic level, when not at optimum levels and exert selection pressure on the genotype for increased adaptation

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## *Genetic Variation & Plant Breeding*

### Characterization of genotypes and environments

- For successful breeding, genotypes should be screened on resistance-susceptibility/ tolerance-sensitivity to the dominant stresses in the future growing area (*=target population of environments*)
- For the target population of environments, the dominant stress factors should be identified and an assessment should be made of their time of occurrence, frequency and intensity



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## *Genetic Variation & Plant Breeding*

### Two approaches to breeding for abiotic stress

- Identify the major stress(es) in the growing area of interest (= target population of environments) and evaluate genotypes in managed stress trials (= trials to which the stress(es) of interest have been applied in a controlled way = selection environments).
- Sample the target population of environments by performing evaluation trials (= selection environments) that are chosen such as to be representative of the target population of environments  
=> multi-environment trials

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## *Genetic Variation & Plant Breeding*

### Multi-environment trial (MET) systems

- Most common approach to abiotic stress breeding is by multi-environment trials = genotypes are evaluated at a number of locations for a number of years
- MET data typically contain many types of genotype by environment interaction
- Trials are *a posteriori* interpreted as having suffered from particular environmental stresses

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## *Genetic Variation & Plant Breeding*

### Examples of GxE in METs

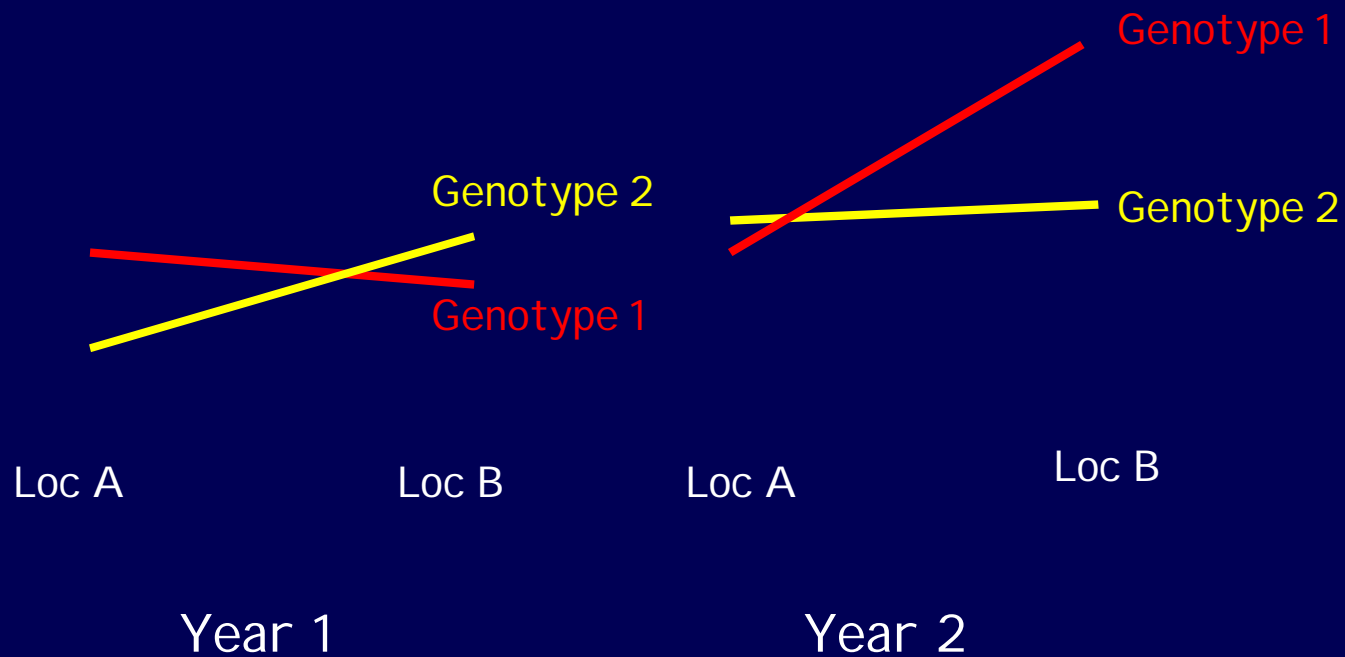
- Genotype x Location = across years, genotype G1 is superior to genotype G2 at location A, but inferior at location B
- Genotype x Year = across locations, G1 is superior to G2 in year 1, but inferior in year 2
- Genotype x Location x Year = in year 1,  $G1 > G2$  at A and  $G1 < G2$  at B; in year 2,  $G1 < G2$  at A and  $G1 > G2$  at B

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## *Genetic Variation & Plant Breeding*

### Examples of GxE in METs (cont)



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## *Genetic Variation & Plant Breeding*

### CIMMYT

#### Wheat program for wide adaptation and drought tolerance

- CIMMYT aims at widely adapted, disease resistant germplasm, high and stable yields across a wide range of environments
- CIMMYT – Basic principles
  - targeted breeding for multi-environments
  - diverse gene pool for crossing
  - shuttle breeding
  - selection for yield under optimum conditions
  - multi-locational testing to identify superior germplasm with good disease resistance

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## *Genetic Variation & Plant Breeding*

### CIMMYT (cont)

#### Targeted breeding-Mega Environment

- To address needs of diverse growing areas CIMMYT introduced concept of mega-environment (ME)
- ME = broad, not necessarily continuous area, occurring in more than one country and frequently transcontinental, defined by similar biotic and abiotic stresses, cropping system requirements, and consumer preferences
- Germplasm developed for a particular ME accommodates the major stresses.

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## *Genetic Variation & Plant Breeding*

### CIMMYT (cont)

#### Shuttle Breeding

- Success of shuttle program depends on
  - Germplasm pool encompassing broad adaptation
  - Selection environments that elicit different response from plant types
- CIMMYT complies with both requirements.
- Advantages of high scale shuttle
  - Higher chance of identifying widely adapted germplasm
  - Easier breaking genetic linkages
  - Easier pyramiding desired genes

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## *Genetic Variation & Plant Breeding*

### CIMMYT (cont)

#### Selection under optimum conditions and breeding for yield potential

- Selection of segregating populations + yield testing of advanced lines is essential for identification of high yielding and input responsive genotypes
- Genetic progress in yield potential related to increase in photosynthetic activity (lower canopy temperature)
- Yield potential seems related to input responsiveness (e.g. N-use-efficiency)



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## *Genetic Variation & Plant Breeding*

### CIMMYT (cont)

#### Durable resistance

- Breeding of widely adapted germplasm with stable yields without adequate resistance against major diseases is impossible. Choice for incorporation of durable, non-specific disease resistance
- Strategy = deployment of historically proven stable genes (non-specific). Accumulate several minor genes, and combine these with specific genes

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## *Genetic Variation & Plant Breeding*

### **ICARDA**

#### Adaptation to low/high input cultivation Ceccarelli - barley program

- Since breeding is mostly conducted under high inputs, opportunities have been missed to exploit genetic differences at low input levels
- Selection should be performed under target levels of input
- Common justification for not selecting under stress conditions is lower  $h^2$  (no experimental evidence for this – Ceccarelli)

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## *Genetic Variation & Plant Breeding*

### ICARDA

#### General arguments

- Adaptation from an agronomic point of view = adapted genotype is able to give an economic production (not merely survive)
- Yield increases achieved in the past are joint outcome of improvement of the agronomic environment and better adapted cultivars
- Strategy based on improvement of the environment may not be sustainable, and is inappropriate for farmers in developing countries
- Adaptation to high input levels can be shown not to extend to low input levels

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## *Genetic Variation & Plant Breeding*

### I CARDA

#### Selection environment and adaptation

- In most crops there is genetic variation for adaptation, and hence yield for low-input conditions
- Most efficient way to improve adaptation and yield in low-input conditions is by direct selection in low-input conditions

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## *Genetic Variation & Plant Breeding*

### I CARDA

#### Selection environment and adaptation

- In most crops there is genetic variation for adaptation, and hence yield for low-input conditions
- Most efficient way to improve adaptation and yield in low-input conditions is by direct selection in low-input conditions

# Seed Production & Quality Management



## *Genetic Variation & Plant Breeding*

### I CARDA

#### General points

- Landraces yield more than modern varieties under low-input
- Superiority is not due to escape mechanisms (see heading date)
- Within landraces there is considerable variation in yield under low input, but they always produce something
- Landraces are responsive to rain and inputs, yield potential of some is high
- Some modern varieties are comparable to landraces under low-input, but this is exceptional

# Seed Production & Quality Management

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## *Genetic Variation & Plant Breeding*

### ICARDA

#### Conclusion

For the choice of selection-environment it is important to know on which side of the cross-over point the target environment is located